

application is unequivocally January 31, 1997. The fluorescent protein embodiment is disclosed and in the possession of the inventor at that time.

Amendments to the Claims are reflected in the listing of claims which begins on page 3 of this paper.

The Remarks/Arguments begin on page 11 of this paper.

-- IN THE CLAIMS --

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

1. (previously presented) A method for identifying an interacting set of proteins comprising:

A) generating first and second fragments of a fluorescent protein reporter molecule which have a directly fluorescent detectable activity when reconstituted and/or associated, wherein said fluorescent protein is selected from the group consisting of green fluorescent protein and mutants of green fluorescent protein;

B) coupling said first fragments of said green fluorescent protein or mutant green fluorescent protein reporter molecule to members of a first panel of proteins;

C) coupling said second fragments of said green fluorescent protein or mutant green fluorescent protein reporter molecule to members of a second panel of proteins;

D) mixing the products of B) and C);

E) directly testing for fluorescence of said green fluorescent protein or mutant green fluorescent protein reporter molecule when reconstituted and/or associated; and

F) identifying the protein panel members whose interaction resulted in fluorescence of said green fluorescent protein or mutant green fluorescent protein reporter molecule and which thus form an interacting set.

2. (previously presented) A method for identifying an interacting set of proteins comprising:

A) identifying a first panel and a second panel of proteins whose mutual interaction is desired to be tested;

B) coupling proteins of said first panel to first fragments of a green fluorescent protein or mutant green fluorescent protein reporter molecule;

C) coupling proteins of said second panel to second fragments of said green fluorescent protein or mutant green fluorescent protein reporter molecule; wherein said first and second fragments have no fluorescent activity prior to step (D)

D) mixing the products of B) and C);

E) directly testing for fluorescent activity of said green fluorescent protein or mutant green fluorescent protein reporter molecule ; and

F) identifying the protein panel members whose interaction resulted in said fluorescent activity and which thus form an interacting set.

3. (previously presented) A method of screening multiple panels of proteins against each other to determine the ability of individual protein panel members to interact with each other, said method comprising:

A) coupling first fragments and second fragments of a green fluorescent protein or mutant green fluorescent protein reporter molecule to different protein panel members; wherein said first and second fragments have no detectable activity;

B) mixing the products of A);

C) testing for said green fluorescent protein or mutant green fluorescent protein reporter molecule activity; and

D) identifying the protein panel members whose interaction results in said fluorescent protein reporter molecule activity and which thus form interacting members.

4. (previously presented) A method according to any of Claims 1-3 where at least two of said panels comprise a library of proteins.

5. (previously presented) A method according to any of Claims 1-3 where at least one of said panels comprises a library of proteins.

6-7. (canceled)

8. (previously presented) A method of preparing an assay system comprising:

A) identifying a first panel of proteins and a second panel of proteins whose mutual interaction is desired to be tested;

B) coupling molecules of said first panel of proteins to first fragments of a green fluorescent protein or mutant green fluorescent protein reporter molecule; and

C) coupling molecules of said second panel of proteins to second fragments of said green fluorescent protein or mutant green fluorescent protein reporter molecule wherein said first and second fragments have no detectable fluorescent activity.

9-10. (canceled)

11. (previously presented) A method for identifying interacting proteins comprising:

(A) generating fragments of a green fluorescent protein or mutant green fluorescent protein reporter molecule, said fragments having a directly detectable activity when associated;

(B) coupling first fragments of said green fluorescent protein or mutant green fluorescent protein reporter molecule to members of a panel of proteins;

(C) coupling a second fragment of said green fluorescent protein or mutant green fluorescent protein reporter molecule to a second protein;

(D) mixing the products of B) and C);

(E) directly testing for said green fluorescent protein or mutant green fluorescent protein reporter molecule activity; and

(F) identifying the panel of protein members whose interaction with said second protein resulted in said fluorescent protein reporter molecule activity.

12. (previously presented) A method for identifying interacting proteins comprising:

(A) identifying a panel of proteins and identifying a second protein whose interaction with members of said panel is desired to be tested;

(B) coupling members of said panel of proteins to first fragments of a green fluorescent protein or mutant green fluorescent protein reporter molecule;

(C) coupling the second protein to a second fragment of said green fluorescent protein or mutant green fluorescent protein reporter molecule wherein said first and second fragments have no detectable fluorescent activity;

(D) mixing the products of B) and C);

(E) directly testing for said green fluorescent protein or mutant green fluorescent protein reporter molecule activity; and

(F) identifying the panel of protein members whose interaction with said second protein resulted in said fluorescent protein reporter molecule activity and which thus form interacting proteins.

13. (previously presented) A method of screening a first protein against a panel of proteins to determine the ability of said first protein to interact with individual members of said panel of proteins comprising:

A) coupling a first fragment of a green fluorescent protein or mutant green fluorescent protein reporter molecule to said first protein;

B) coupling second fragments of said green fluorescent protein or mutant green fluorescent protein reporter molecule to different members of said panel of proteins wherein said first and second fragments have no detectable fluorescent activity;

C) mixing the products of A) and B);

D) testing for fluorescent activity of said green fluorescent protein or mutant green fluorescent protein reporter molecule; and

E) identifying the members of said panel of proteins whose interaction with said first

protein results in said fluorescent protein reporter molecule fluorescent activity and which thus interact with said first protein.

14. (previously presented) A method according to any of Claims 11-13 wherein said panel comprises a library of proteins.

15-16. (canceled)

17. (previously presented) A method of preparing an assay system comprising: (A) identifying a panel of proteins whose interactions with a second protein are desired to be tested; (B) coupling members of said panel of proteins to first fragments of a green fluorescent protein or mutant green fluorescent protein reporter molecule; and (C) coupling said second protein to a second fragment of said green fluorescent protein or mutant green fluorescent protein reporter molecule wherein said first and second fragments have no detectable fluorescent activity.

18-19. (canceled)

20. (previously presented) A method for identifying interacting proteins comprising: (A) generating fragments of a green fluorescent protein or mutant green fluorescent protein reporter molecule which have a directly fluorescent detectable activity when associated; (B) coupling a first fragment of said green fluorescent protein or mutant green fluorescent protein reporter molecule to a first protein; (C) coupling a second fragment of said green fluorescent protein or mutant green

fluorescent protein reporter molecule to a second protein; (D) mixing the products of B) and C); and (E) directly testing for fluorescent activity of said fluorescent protein reporter molecule in the absence or presence of one or more chemical or biological compounds.

21. (previously presented) A method for identifying interacting proteins comprising:

- A) identifying a first protein and a second protein whose interaction is desired to be tested;
- B) coupling said first protein to a first fragment of a green fluorescent protein or mutant green fluorescent protein reporter molecule;
- C) coupling said second protein to a second fragment of said green fluorescent protein or mutant green fluorescent protein reporter molecule wherein said first and second fragments have no detectable fluorescent activity;
- D) mixing the products of B) and C);
- E) directly testing for fluorescent activity of said fluorescent protein reporter molecule.

22-23 (canceled)

24. (previously presented) A method of preparing an assay system comprising:

- A) identifying a first protein and a second protein whose interaction is desired to be tested;
- B) coupling said first protein to a first fragment of a green fluorescent protein or mutant green fluorescent protein reporter molecule; and

C) coupling said second protein to a second fragment of said green fluorescent protein or mutant green fluorescent protein reporter molecule wherein said first and second fragments have no detectable fluorescent activity.

25-29. (canceled)

30. (previously presented) A method according to any of Claims 1-3, 8, 11-13, 17, 20-21, and 24 wherein said green fluorescent protein reporter molecule generates an optically detectable signal.

31. (previously presented) A method according to any of Claims 1-3, 8, 11-13, 17, 20-21, and 24 wherein said reporter molecule generates a fluorescent signal.

32. (previously presented) A method according to any of Claims 1-3, 8, 11-13, 17, 20-21, and 24 wherein said fluorescent protein reporter molecule generates a signal that can be quantified within living cells.

33. (previously presented) A method according to any of Claims 1-3, 8, 11-13, 17, 20-21, and 24 wherein said fluorescent protein reporter molecule generates a signal that can be localized within living cells.

34 - 36 (canceled)

37. (currently amended) A method according to any of Claims 1-3, 8, 11-13, ~~14~~, 17, 20-21, and 24 wherein the fluorescent protein reporter molecule activity is detected by one or more methods selected from the group consisting of: cell color, fluorescence, optical density, spectroscopy, flow cytometry, microscopy, or image analysis.